IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants : Hiroshi SUSAKI et al.

Group Art Unit : 1646

Appl. No. : 10/822,661

Examiner : Russel

Filed : April 13, 2004

Confirmation No. : 3663

For : DDS COMPOUND AND METHOD FOR MEASUREMENT THEREOF

**DECLARATION UNDER 37 C.F.R. 1.132 OF YOSHINOBU SHIOSE**

Commissioner for Patents
U.S. Patent and Trademark Office
Customer Service Window, Mail Stop _____
Randolph Building
401 Dulany Street
Alexandria, VA 22314

Sir:

I, Yoshinobu SHIOSE, being duly warned, declare that:

1. I am a citizen of Japan, and have been employed by Daiichi Pharmaceutical Co., Ltd., since April, 1991.

2. I graduated from Gifu Pharmaceutical University in March 1989, and received a Master's Degree from Gifu Pharmaceutical University in March, 1991.

3. I have been involved in research relating to antitumor agents, and isolation and purification of new pharmacologically-active substances, and research relating to drug delivery system compounds having a polymer moiety.

4. Presently, I am Associate Senior Researcher in Drug Metabolism & Physicochemical Property Research Laboratory of Daiichi Pharmaceutical Co., Ltd.

5. I am familiar with the invention of U.S. Patent Application No. 10/822,661 and its parent application, i.e., Application No. 09/807,980, now U.S. Patent No. 6,811,996.

6. It has been brought to my attention that in the above parent application, i.e., Application No. 09/807,980, the Examiner has rejected claims similar to those pending in the 10/822,661 application in an Office Action mailed April 17, 2003.

In particular, the rejection asserts, amongst other assertions, that:

> It would have been obvious to one of ordinary skill in the art at the time Applicants' invention was made to attach galactose or mannose residues to the drug complexes of the WO Patent Application '260 comprising the drug of Applicants' claim 19 because the Japanese Patent Application '746 discloses that the drug is useful in treating cancer of the liver and because Theodore et al, the Gonsho et al article, the Hashida et al article, the Kichler et al article, and the Nishikawa et al article teach that attachment of various galactose or mannose residues to polymeric drug carriers is a known and conventional method for targeting drugs to the organ to be treated.

7. The following tests have been conducted under my supervision utilizing a galactose-modified compound, a compound without galactose modification and a control antitumor agent.

8. In particular, the following compounds were tested:

(a) A compound according to Applicants' invention (galactose-modified CM-dextran polyalcohol-Gly-Gly-Phe-Gly-DX-8951) was prepared according to the methods of Example 6(A) to 6(C) disclosed in the specification of Application No. 10/822,661 and its parent Application No. 09/807,980 (hereinafter also referred to as "Compound 8(a)").

(b) A comparative compound (CM-dextran polyalcohol-Gly-Gly-Phe-Gly-DX-8951 not modified with galactose) was prepared according to the method of Example 6(D) disclosed in the specification of Application No. 10/822,661 and its parent Application No. 09/807,980 (hereinafter also referred to as "Comparative Compound 8(b)").

(c) DX-8951 which is an antitumor agent per se, which was utilized as a control substance.

9. The following experimental method was followed:

M5076 cells (murine histiocytoma) were intravenously transplanted into male C57BL/6 mice ($1 \times 10^5$ cells/mouse). Eight days after the transplantation, the Compound 8(a) of the present invention (galactose-modified) or the Comparative Compound 8(b) (non-modified) was intravenously administered to the mice at 10, 20,40, or 60 mg/kg (dose levels expressed as DX-8951, each group consisting of 10 mice). The effect on prolongation of survival period was studied. Survival time was monitored until termination of the study on day 70, and dead mice were necropsied and the presence or absence of tumor tissue in every organ of them was visually inspected. After 70 days from the transplantation, surviving mice were sacrificed and autopsied, and subjected to the same inspection. The life prolongation effect was indicated as increase in life span (ILS) calculated from the following formula:

$$ILS(\%) = [(MSTt)/(MSTc) - 1] \times 100,$$

wherein MSTt and MSTc represent the median survival time of a treated group in days and that of the control group in days, respectively.

10. Table 1 below shows experimental results of antitumor effects of Compound

8(a) with galactose modification and Comparative Compound 8(b) without galactose modification in a liver metastasis model of murine histiocytoma M5076.

Table 1

Antitumor effect of galactose-modified Compound 8(a) and Compound 8(b) not modified with galactose in a liver metastasis model of murine histiocytoma M5076 (intravenous transplantation)

| Compound | Dose (mg/kg) | Administration day | Survival days Range | MST | | | ILS(%) | BWLmax[a] (%)[Day] | N[b] | S[c] |
|---|---|---|---|---|---|---|---|---|---|---|
| Control | 0 | - | 14-16 | 15.14 | | | 0.0 | 4.6[15] | 0/10 | - |
| DX-8951 (antitumor agent, | 60 | 8 | 22->70 | 34.75 | ** | | 129.5 | 4.5[12] | 0/10 | 0/1 |
| per se) | 20 | 8 | 18-21 | 19.00 | ** | | 25.5 | 2.7[9] | 0/10 | - |
| Galactose-Modified | 60 | 8 | 32->70 | 50.00 | ** | | 230.3 | 6.3[54] | 0/10 | 1/1 |
| Compound 8(a) | 40 | 8 | 27-66 | 47.00 | ** | ¬ | 210.4 | 2.1[9] | 0/10 | - |
| CM(0.7)·Dex·PA(t-Gal)· | 20 | 8 | 19-27 | 20.33 | ** | \| ## | 34.3 | 1.8[9] | 0/10 | - |
| GGFG·DX·8951 | 10 | 8 | 17-19 | 17.80 | | \| | 17.6 | 2.8[18] | 0/10 | - |
| (MW of carrier: 14K) | | | | | | | | | | |
| Without Galactose | 60 | 8 | 26->70 | 39.25 | ** | \| | 159.2 | 2.2[9] | 0/10 | 1/2 |
| Modification | 40 | 8 | 23-29 | 28.00 | ** | _\| | 84.9 | 2.9[9] | 0/10 | - |
| Compound 8(b) | 20 | 8 | 18-20 | 18.67 | ** | | 23.3 | 2.2[9] | 0/10 | - |
| CM(0.7) ·Dex·PA·GGFG· | 10 | 8 | 17-19 | 17.42 | | | 15.1 | 1.7[7] | 0/10 | - |
| DX-8951 | | | | | | | | | | |
| (MW of carrier: 14K) | | | | | | | | | | |

[a] Maximum value for rate of body weight loss (%), numbers in brackets denote the day.
[b] Number of mice that died of toxicity/number of mice used.
[c] Number of mice without tumor tissues or nodules/number of mice survived on day 70 after tumor inoculation.
The dose levels of the compounds are expressed as those of DX-8951.
** $P \leqq 0.01$, * $P \leqq 0.05$ versus control, ## $P \leqq 0.01$ by Williams·Wilcoxon test.

11. From the experimental results, it can be understood that the mouse group administered with Compound 8(a) with galactose modification gave longer survival date with significant difference (marked with "##" on the comparison of the values 47.00 and 28.00 in Table 1 that means $P \leq 0.01$ by Williams-Wilcoxon test). This advantageous antitumor effect of Compound 8(a) with galactose modification, which is significantly higher than the antitumor effect of Comparative Compound 8(b) without galactose modification, is believed to be attributable to accumulation of the galactose modified Compound 8(a) to the liver with the aid of the galactose modification.

12. A prior publication of Duncan et al., Journal of Controlled Release, 19 (1992) 331-346 (hereinafter "Duncan"), discloses a result of antitumor effect of a compound in which doxorubicin is bound with polymer. Table 3 of Duncan (on page 339) discloses experimental results obtained by employing a similar experimental system that was used in the experiments discussed in paragraph 9 above. In Table 3 of Duncan, prolongation effects on survival days by administration of Polymer 1 (without saccharide modification) and that of Polymer 2 (with saccharide modification, whose chemical structure is shown in Fig. 1 of Duncan, page 332) are compared. Polymer 1 without saccharide modification gave a higher number of surviving mice, which means that this compound has higher antitumor effect than Polymer 2 that has saccharide modification. Accordingly, on information and belief, one having ordinary skill in the art would have recognized that before Applicants' invention, that saccharide modification of a conjugate compound of an antitumor agent bound with a polymer would not yield an improvement of antitumor effect of the conjugate compound. Therefore, one of ordinary skill in the art

would not have expected the advantageously high antitumor effect against metastatic liver cancer according to Applicants' application.

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true, and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Yoshinobu Shiose
Yoshinobu SHIOSE

February 4, 2005
Date